Short Communication

Effect of dual inoculation with *Rhizobium* and vesicular arbuscular mycorrhiza in chickpea (*Cicer arietinum*) at varying nitrogen levels

Poonam Sharma* and A. S. Khurana
Department of Plant Breeding, Punjab Agricultural University, Ludhiana 141 004, India.

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ABSTRACT

Field experiments were conducted for two consecutive years (*rabi* 1995-96 and 1996-97) to study the effect of various levels of N (0, 25, 50 and 75 kg ha^{-1}) with *Rhizobium* alone and in combination with vesicular arbuscular mycorrhiza (VAM) on nitrogen fixation, VAM colonization and grain yield in chickpea. Maximum number and dry weight of nodules and nitrogenase activity were found at 50 kg N ha^{-1} in dual inoculated treatments. Nodulation and nitrogenase activity were inhibited at 75 kg N ha $^{-1}$. However, maximum percentage of mycorrhizal colonization and grain yield were recorded at 75 kg N ha^{-1}. Thus, higher level of N might facilitate VAM cononization and increased grain yield of chickpea in the presence of *Rhizobium* inoculation.

Keywords : Chickpea, *Cicer arietinum* (L.), *Rhizobium*, nitrogenase activity, vesicular arbuscular mycorrhiza, nitrogen, India

The beneficial effect of *Rhizobium* inoculation with vesicular-arbuscular mycorrhiza (VAM) on nutrient uptake, particularly phosphorus is well documented, which in turn is known to stimulate biological nitrogen fixation (Subba Rao *et al.* 1982). The information on the effects of various levels of nitrogen on VAM colonization is limited and contradictory (Azcon *et al.* 1982; Thompson 1986; Tilak and Dwivedi 1991). Present work was therefore, undertaken to study the effect of various levels of N on VAM colonization, nitrogen fixation and grain yield in field grown chickpea.

Field experiments were conducted in microplots (1.5 m x 1.2 m) for two consecutive years durirng *rabi* (October-April) 1995-96 and 1996-97 at the experimental farm, Punjab Agricultural University, Ludhiana in the subtropical region of India, to study the effect of various levels of N on VAM colonization, nitrogen fixation and grain yield in chickpea variety PBG 1. The soil was a loamy sand, low in organic carbon (0.32%) and available nitrogen (103 kg ha^{-1}), medium in available phosphorus (14.5 kg ha^{-1}) and potassium (178 kg ha^{-1}) with pH 8.2. A basal dose of 40 kg P_2O_5 ha^{-1} was applied at sowing. Chickpea seeds were coated with recommended culture of *Rhizobium* (PAU G-33) in all treatments except the uninoculated control. There were four treatments at nitrogen rates of 0, 25, 50 and 75 kg ha^{-1} in the form of urea. Four microplots consisting of different levels of N were inoculated with *Rhizobium* and sand + soil (1:1) based inoculum of *Glomus fasciculatum* (Thaxter sensu Gerrd.) Gerrd. & Trappe. Two hundred and fifty grams of inoculum (125-150 spores per 100 g) were spread uniformly in four microplots.

Five plants were carefully uprooted from each plot at 65 days after sowing and gently washed to record nodulation. The nodules were detached and oven dried at 60^0 C for 24 hours and the dry weight was recorded. The nitrogenase activity of nodules with intact root system was measured by acetylene reduction assay by gas chromatography (AMIL NUCON MODEL 5560) using porapak-R column (Hardy *et al.* 1968) and percent colonization was determined according to Phillips and Hayman (1970). Grain yield was recorded at the harvesting stage. The experiment was laid out in a Randomized Block Design with four replications. The data were subjected to analysis of variance and the mean seperation was done by least significance difference test.

Higher number and nodule dry weight/plant and nitrogenase activity were recorded in dual inoculated treatmen at all N levels. Maximum number, dry weight and nitrogenase activity of nodules (48, 74 mg and 90.5 μ moles of C_2H_4 hr^{-1}plant^{-1}) were recorded at 50 kg N ha^{-1} in dual inoculated treatments. Although nodulation and nitrogenase activity decreased, significant percentage of VAM colonization and grain yield were recorded at 75 kg N ha^{-1} i.e. 75.5% and 1680 kg ha^{-1} were recorded at this level (Table 2).

Table 1. Number, dry weight and nitrogenase activity of nodules as influenced by inoculation with VAM and Rhisobium at varying levels of N (Pooled analysis)

Treatment	Number of Nodules Plant^{-1}	Nodule dry wt. plant^{-1}(mg)	Nitrogenase activity (Um of $C_2H_4h^{-1}Plant^{-1}$)
Uninoculated Control	20	38 0	40.5
0Kg N ha^{-1}			
Rhisobium ®	22	45.5	48.5
R+VAM	25	51.0	55.5
25 Kg N ha^{-1}			
R	28	55.0	68.0
R+VAM	33	60.0	74.5
50 Kg N ha^{-1}			
R	37	65.5	77.0
R+VAM	48	74.0	90.5
75 Kg N ha^{-1}			
R	35	62.0	70.5
R+VAM	44	70.5	85.2
C.D 5%	4.2	12.5	20.8

Table 2. Root colonization and grain yield as influenced by inoculation with VAM and *Rhizobium* at varying levels of N (Pooled analysis)

Treatments	Root colonization (%)	Grain Yield (kg ha^{-1})
Uninoculated Control	22.0	1455
0 Kg N ha-1		
Rhizobium (R)	28.0	1470
R+VAM	35.0	1565
25 Kg N ha-1		
R	37.5	1490
R+VAM	55.0	1605
50 Kg N ha-1		
R	45.0	1505
R+VAM	60.0	1640
75 Kg N ha-1		
R	52.0	1530
R+VAM	75.5	1680
C.D. 5%	7.5	100.5

Tilak and Dwivedi (1991) also reported significant increase in VAM colonization and grain yield in chickpea at 100 kg N ha^{-1}.

It has been reported that higher levels of N might facilitate VAM colonization (Thompson 1986; Kumar *et al.* 1998 and Konde *et al.* 1998) and subsequently increase grain yield of chickpea in the presence of *Rhizobium* inoculation.

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Short Communication

Tomato production using chemical fertilizer and nasute termite mound as a soil amendment in Nigeria

E.C. Nzegbule
College of Natural Resources and Environmental Management, Federal University of Agriculture, Umudike, P.M.B.7267 Umuahia, Nigeria. E-mail: nzegbule@yahoo.com

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ABSTRACT

A local soil amendment, nasute termite mound, was compared with chemical fertilizer ($N.P_2O_5K_2O$:15:15:15) for its ability to improve tomato (*Lycopersicum esculentum* L.) yield. Tomato seedlings were grown on top soil only, top soil fertilized with NPK and a 3:7 kg crushed top soil mixture. The mound had significantly higher percentages of clay and silt contents. The chemical analysis indicated that the total N% organic C%, available P (ppm), and effective CEC (Cmol(+) kg^{-1}), were increased by 33.6%, 47.8%, 34%, and 20.1% respecting in crushed mound over top soil. Crushed mound-top soil mixture significantly increased fruit yield and plant dry weight of tomato compared to top soil only. Fruit yields were 95, 88.6 and 54.5g per pot for crushed mound-top soil mixture, NPK and top soil only, respectively. Total dry matter of tomato grown on NPK fertilizer mixed with top soil was 5.8% which was significantly lower than that of crushed mound-top soil mixture. Crushed mound-top soil mixture and NPK fertilized top soil induced similar improvement of fruit and plant dry weight of tomato. However, production of tomato with crushed mound-top soil mixture had greater Output-Input ratio than using chemical fertilizers. Hence crushed nasute termite mound could be suggested as a cheap and effective soil amendment for resource-poor farmers.

The traditional bush-fallow system which was highly dependent on availability of sufficient land collapsed because of rapid rates of population growth and competition for land in Nigeria (IITA 1992). In recent years increased demand for food and pressure on land led to the replacement of traditional agriculture with intensive cultivation, which in turn caused poor crop yield and degradation of soil (Ofori 1993). This problem was aggravated by low use of inputs due to farmers' inability to purchase required material such as fertilizers. The resource-poor farmers became more impoverished as the returns from land no longer compensated for cost of labour and other production cost. This emphasizes the need to identify effective and low cost sources of locally available inputs (Nzegbule and Osodeke 1997). An earlier survey showed that termite mounds were commonly found in forest and fallow lands in Southern Nigeria, with a mean density of 76 mounds per hectare. These mounds (termiteria) were mushroom shaped and were on average 0.66m in height with about 0.39 m basal circumference. Nasute termite mounds are dark brown in colour and have sticky and plastic consistence when wet and become very hard when dry. The fertility value of this material has been associated with soil activities of termites as decomposers (White 1979; Anon 1996). Although the potential of nasute termite mounds to enhance crop yield has been established (Nzegbule and Osodeke 1997), there is a need to compare yield enhancing capacity of the termite mound with that of NPK chemical fertilizer.

Tomato (CV. U 143-0-4B-1-0-0) seeds were planted in pots filled with 10 kg of either top soil or a crushed mound-top soil mixture. The ratio of crushed mound and top soil was 3:7 kg. Samples of nasute termite mound and top soil were separately oven-dried and ground for mechanical and chemical analysis at the soil laboratory, Federal University of Agriculture, Umudike, Nigeria. A sub-sample was digested for determination of total N content using micro-kjeldahl method (Jackson 1962). Organic C was determined by the Walkey - Black method (Allison 1965), K and Na (Cmol(+)kg^{-1}) by the flame emission photometry (Black 1965), available P content by Bray method 1 (Bray and Kurt 1945) and Ca and Mg (Cmol(+) kg^{-1}) by the ethylene diaminetetra -acetic acid (EDTA). The mechanical analysis was done using the procedure of Bouyoucos (1951). At germination the seedlings were thinned to one per pot. Tomato seedlings grown with chemical fertilizer were added with 8g of $N.P_2O_5.K_2O$ (15:15:15) at 10 days after germination. Another set of seedlings was grown on top soil only and these were used as the control. A completely randomized design with five replications was used in conducting the trial in a glasshouse at the Federal University of Agriculture, Umudike, Nigeria. At maturity tomato fruits and plant dry matter were harvested and weighed. Data collected on soil parameters, tomato fruit yield and plant dry weight were statistically analyzed using student t-test and Fisher's least significant difference (LSD) (Steel and Torrie 1980). Also an Output- Input analysis was used to evaluate the profitability of crushed mound to produce tomato.

Table 1. Physical and Chemical composition of nasute termite mound and topsoil in Umudike, Nigeria.

Composite	Termite mound	Top soil (0-15 cm)
Clay %	21.40*	13.4
Silt %	6.90*	2.0
Sand %	71.70*	84.6
PH (H_2O)	5.45	4.92
Total N %	0.29	0.144
Organic C %	3.12	1.10
C/N	10.4*	7.63
Available P (ppm)	9.35*	4.6
Exch. K (Cmol(+) kg^{-1})	1.38*	0.28
Exch.Na (Cmol(+) kg^{-1})	0.37*	0.18
Exch. Ca (Cmol(+) kg^{-1})	0.84	0.70
Exch. Mg (Cmol(+) kg^{-1})	0.31	0.59
Exch. Acidity (Cmol(+) kg^{-1})	1.19	0.96
Effective CEC (Cmol(+) kg^{-1})	4.08*	2.71

* = significantly different at 5% along the row.

The clay and silt content of nasute termite mound were 38.9% and 55%, respectively which was higher than that of top soil (Table 1). The acid sandy soils of Southern Nigeria are characterized by coarse texture, poor water holding capacity and high rate of leaching. Hence the incorporation of crushed mound will improve soil physical properties through the increase of clay and silt content (Agboola and Obatolu 1989). Soils with higher activity clay minerals are known to have reduced rate of leaching and improved moisture holding capacity. The total N%, Organic C%, available P (ppm), K, Na and Ca were higher by 33.6%, 45.8%, 34%, 67%, 34.2% respectively in crushed mound than the top soil. Evidently, nasute termite mound has a high fertility level as a result of the base materials on which the termites feed on and construct their mounds (Anon 1996). When crushed mound-top soil mixture and N.P.K fertilized topsoil were separately used to grow tomato, the fruit and plant dry matter yields were significantly higher than the control by 42% and 35% respectively. The low fruit yield and plant dry weight of tomato grown on top soil indicated the state of depletion of soil fertility as reported in Table 1.

Table 2. Effect of crushed mound - top soil mixture and chemical fertilizer (NPK) on fruit yield and plant dry weight of tomato.

Medium	Fruit yield ($g\ pot^{-1}$)	Plant dry weight ($g\ pot^{-1}$)
Crushed mound-Topsoil	95.0	22.8
NPK-Top soil	88.6	24.6
Top soil	54.5	19.6
LSD	15.92	2.61

Igbokwe *et al.* (1982) have noted that depletion of soil fertility in the area was mainly caused by continuous cultivation and nutrient leaching. Continuous cultivation practised by farmers in the area accelerated export of soil nutrients in the form of harvested crops (Zake 1993).

Maximum fruit yield (95 g pot^{-1}) was produced when crushed mound -top mixture was used, and this was 6.7% higher than that of NPK fertilized top soil. Also, the plant dry weight of tomato was 24.6 g pot^{-1} when grown with NPK and 22.8 g pot^{-1} when grown with crushed mound-top soil mixture (Table 2). Considering the parameters measured, the performance of tomato when grown on crushed mound-top soil mixture was not markedly different from that of chemical fertilizer. Perhaps, the incorporation of crushed mound improved the nutrient availability of the top soil in the area which is high in acidity, low in organic matter and other soil nutrients (Agboola and Obatolu 1989).The crushed mound which has been noted to have high percentage of clay, pH, total N% organic C% and available P% may have induced greater fruit yield and plant dry

Table 3. Output-input analysis for the production of 100 potted stands of tomato using crushed mound and chemical fertilizer.

Variable	Quantity	Unit cost (US$)*	Amount (US$)*
(A) Input			
Acquisition Cost			
Mound	700kg	0.00	0.00
Fertilizer (NPK)	0.8 g	0.3 g	6.00
Cost of Topsoil used			
Mound	300 kg	1.50 kg	6.00
Fertilizer	700 kg	1.50 kg	14.00
Transportation cost			
Mound (from farm)	700 kg	1.5 (1 Monday)	1.5
Fertilizer (from market)	0.8 kg	1.0	1.00
Application cost			
Mound (crushing/mixing)	700 kg	1.5 day	300
Fertilizer	0.8 kg	1.5 day	1.5
Total Input cost			
Mound	-	-	10.5
Fertilizer	-	-	16.75
(B) Output			
Fruit yield			
Mound	9.5 kg	2 kg	19.00
Fertilizer	8.9 kg	2 kg	17.8

*US $ 1 = N100 Nigerian currency.

weight of tomato by ameliorating the nutrient content of the top soil.

The economic viability of producing tomato using crushed mound-top soil mixture was apparently clear as could be seen from the assessment of the Output- Input analysis (Table 3). The output-input ratio for growing tomato using crushed mound was 1.8:1 while that of chemical fertilizer was 1.1:1.

Therefore use of crushed mound-top soil mixture has a greater efficiency than chemical fertilizer on the basis of cost of output- input analysis.

Nasute termite mound when incorporated with top soil promoted fruit yield and plant dry weight of tomato just as when chemical fertilizer (N.P.K) was used. Considering the advantages termite mound has over chemical fertilizers, in terms of ease of availability and economic benefits, it could be useful as a cheap soil amendment for tomato and other vegetable crop production by local farmers.

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